

TWO TYPES OF TRICHROMATIC SQUIRREL MONKEY SHARE A PIGMENT IN THE RED-GREEN SPECTRAL REGION

J. K. BOWMAKER,¹ G. H. JACOBS,² D. J. SPIEGELHALTER³ and J. D. MOLLON⁴

¹School of Biological Sciences, Queen Mary College, University of London, Mile End Rd,
London E1 4NS, England

²Department of Psychology, University of California, Santa Barbara, CA 93106, U.S.A.

³MRC Biostatistics Unit, Medical Research Council Centre, Hills Rd, Cambridge CB2 2QH, England

⁴Department of Experimental Psychology, University of Cambridge, Downing St., Cambridge CB2 3EB,
England

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Abstract—Microspectrophotometric measurements have been obtained for individual photoreceptors from four female squirrel monkeys (*Saimiri sciureus*) that had been shown behaviourally to be trichromatic. Relative to a normal human observer, two of the monkeys required more red light for a Rayleigh match; the other two required more green light than a normal human observer. In the red-green spectral region, the first type of monkey was found to have two cone pigments with peak sensitivities at approximately 536 and 549 nm, whereas the second type was found to have pigments with peak sensitivities at approximately 549 and 564 nm. By maximum likelihood estimation it was shown that the microspectrophotometric data could be described by a model that assumed only three underlying distributions, two of which were present in each type of monkey. The fit of this model was as good as one in which a "double normal" distribution was fitted individually to the data for each animal. This result is consistent with a genetic theory that postulates in *Saimiri* three possible alleles for a single locus on the X-chromosome; the heterozygous female enjoys trichromacy because Lyonisation ensures that only one photopigment is manufactured in any given cone.

Colour vision Microspectrophotometry Visual pigments Genetics Anomalous trichromacy
Squirrel monkeys *Saimiri sciureus*

INTRODUCTION

The squirrel monkey (*Saimiri sciureus*) exhibits a remarkable polymorphism of colour vision. Amongst the members of this species both trichromatic and dichromatic individuals are found; and within each of these two broad classes there appear to be several distinguishable sub-types (Jacobs, 1984). All the males that have been tested have proved to be dichromatic, whereas both dichromatic and trichromatic females have been found.

In a recent double-blind study, microspectrophotometric measurements were obtained for samples of photoreceptors from ten individual monkeys, whose colour vision had in each case been tested behaviourally (Mollon *et al.*, 1984). The pooled microspectrophotometric records suggested that at least four types of cone pigment could be found in this species and that any individual monkey could draw either two or three pigments from the total set. A short-wave cone pigment, with a wavelength of peak absorbance (λ_{\max}) at about 433 nm, was thought to be common to all individuals. In the middle-to-long wave part of the spectrum there appeared to be at least three possible cone pigments; and it was in this region that the number and spectral positions of the photopigments varied between individuals. Animals that had been behaviourally classified as dichromatic had only one photopigment in the middle-to-long

wavelength part of the spectrum, whereas behaviourally trichromatic animals appeared to have two photopigments in this spectral region. Two types of trichromat were present in the sample: one type behaviourally resembled protanomalous human observers and the other type resembled deuteranomalous human observers.

The present study concerns squirrel monkeys of the trichromatic type and was undertaken to answer one question that was not satisfactorily answered by the earlier study: do "protanomalous" and "deuteranomalous" monkeys share a common pigment with λ_{\max} close to 550 nm? The earlier sample included three "deuteranomalous" monkeys, and in the middle- to long-wave part of the spectrum the collected λ_{\max} values for individual cones from these animals were bimodally distributed, with clear peaks near 550 and 565 nm. However, there was only one "protanomalous" monkey in the sample and the distribution of λ_{\max} values for this monkey, though broad and significantly non-normal, could not be subdivided with confidence; thus, it was not possible to estimate precisely the λ_{\max} values of the two photopigments suspected to underlie the distribution. We have therefore obtained behavioural and microspectrophotometric results for four more trichromatic squirrel monkeys. A statistical analysis of the microspectrophotometric records has been designed

to answer the specific question of whether "protanomalous" and "deuteranomalous" squirrel monkeys share a common pigment in the green-yellow part of the spectrum.

The question asked here has also been raised with respect to human anomalous trichromats (see e.g. De Vries, 1948; MacLeod and Hayhoe, 1974; Pokorny and Smith, 1977), but the question has a particular interest of its own in the case of the squirrel monkey. The results of previous studies (Mollon *et al.*, 1984; Jacobs and Neitz, 1985) led to a genetic model that requires the existence of pigments in the red-green range, that are common to more than one type of trichromatic squirrel monkey. The model proposes that in *Saimiri*—unlike man—there is only one genetic locus for generating a pigment in the red-green range, that this locus is on the X chromosome, and that at least three alleles can occur at the locus. A female squirrel monkey gains trichromacy if she inherits two different alleles at this locus, whereas her male conspecifics, having only one X chromosome, can never be other than dichromats. As it stands, this model requires that any individual female can randomly draw two alleles from the available set; and thus there should exist pigments in the red-green range that are shared by more than one phenotype.

METHODS

The behavioural and microspectrophotometric measurements were made independently, the former in Santa Barbara, California, and the latter in London. Details of the behavioural results for a given monkey were not known to the microspectrophotometrists while they were making their measurements.

Subjects

The subjects were four female squirrel monkeys (*Saimiri sciureus*). Two were of the Roman Arch variety, two of the Gothic Arch variety (MacLean, 1964; Cooper, 1968). The former are here identified by their laboratory numbers as S26 and S36, the latter as S11 and S14.

Behavioural measurements

Before the microspectrophotometric measurements, each animal was tested behaviourally to establish the nature of its colour vision. The apparatus and procedure used for the behavioural measurements have been described by Jacobs (1983; 1984). The animals made forced-choice discriminations: they viewed three transilluminated panels and were reinforced for selecting the panel that appeared different from the other two. Results are reported for tests of three types: (i) Increment-threshold measurements for 540-nm and 640-nm test lights, which were superimposed on continuously present achromatic fields (luminance = 3.4 cd/m⁻²); (ii) wavelength-discrimination thresholds measured

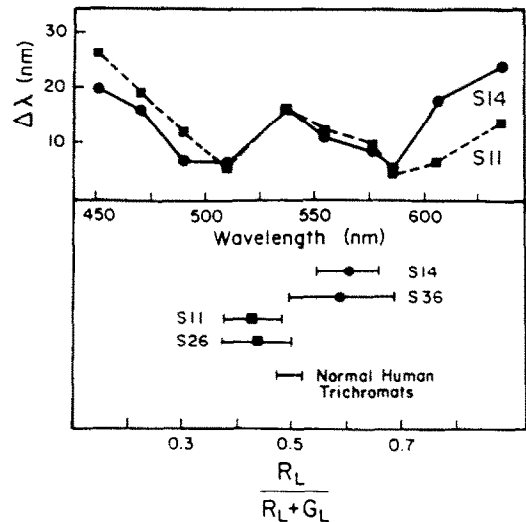


Fig. 1. Results of behavioural tests. The upper panel shows wavelength-discrimination functions for two individual animals: the ordinate represents the magnitude of the wavelength difference required at each spectral location to support discrimination at the criterion level of 70% correct. The values plotted are the averages for threshold differences in both spectral directions, except for the two extreme wavelengths tested; in the latter cases, discrimination could be measured only for test values lying closer to the middle of the spectrum. The lower panel shows Rayleigh matches for the four individual animals: the value plotted is the midpoint of the red/green mixture range over which discrimination did not differ significantly from chance. The horizontal bar shows the entire range of matches for 40 normal human trichromats.

at ten spectral locations between 450 and 630 nm; (iii) a Rayleigh-match test, in which monkeys were required to discriminate various mixtures of green (536 nm) and red (625 nm) lights from a yellow (585 nm).

Microspectrophotometric measurements

The measurements in London were made with a dual-beam Liebman microspectrophotometer (Liebman and Entine, 1964; Knowles and Dartnall, 1977, pp. 562–566), which had been modified and placed under the control of a laboratory computer. Details of the procedures are given by Bowmaker *et al.* (1980) and Mollon *et al.* (1984). Under infra-red inspection, the measuring beam of the microspectrophotometer was arranged to pass through a single outer segment while a reference beam passed through adjacent clear space in the preparation. The measuring beam usually passed transversely through an outer segment, but we sometimes made near-axial measurements when we came upon a matrix of receptors lying perpendicular to the plane of the microscope stage.

The primary data for an individual cell consisted of a plot of absorbance (optical density) as a function of wavelength. From such a record we derived an estimate of λ_{max} as follows. First, the absorbance values at pairs of adjacent wavelengths were averaged to obtain a mean curve from the outward and return

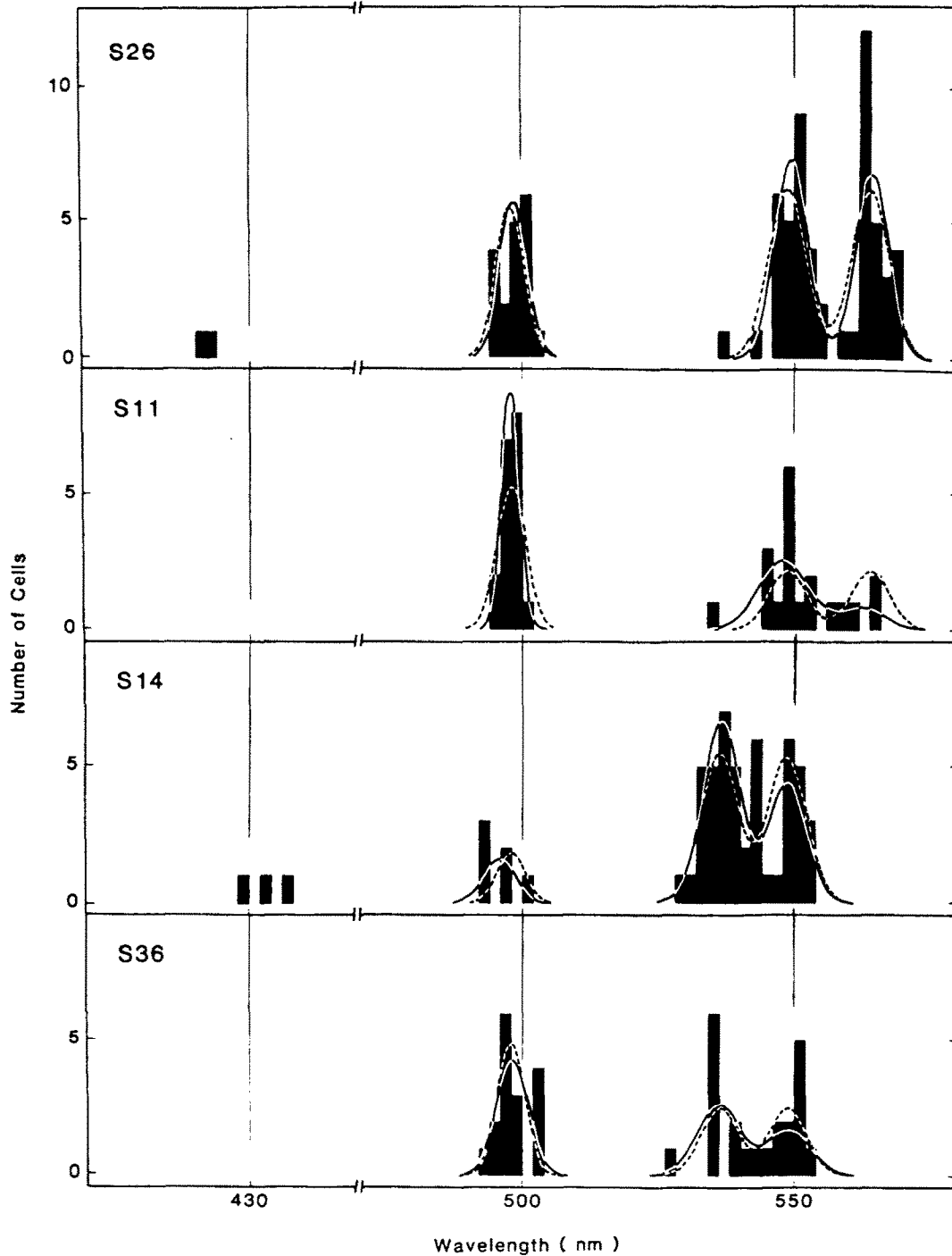


Fig. 2. Distribution of values of peak sensitivity (λ_{max}) of individual microspectrophotometric records. Distributions are shown separately for each monkey. The upper two plots show results for the monkeys that were behaviourally deuteranomalous; the lower two plots show results for the monkeys that were behaviourally protanomalous. The continuous lines represent the best-fitting distributions when the fits are obtained independently for each monkey (Model 1; see text); the dashed lines represent the best-fitting distributions obtained when λ_{max} values and standard deviations are constrained to be consistent across animals (Model 2; see text).

traces. Each of 20 absorbance values on the long-wavelength limb of the curve (corresponding to a 40-nm segment of the trace and to absorbances in the range of approximately 45–90% of the maximum for that cell) was then referred to a standard template

curve to obtain an estimate of the λ_{max} ; this operation amounts to finding the spectral location of the standard curve that gives the percentage absorbance value under consideration. The 20 individual estimates were then averaged to give the values entered

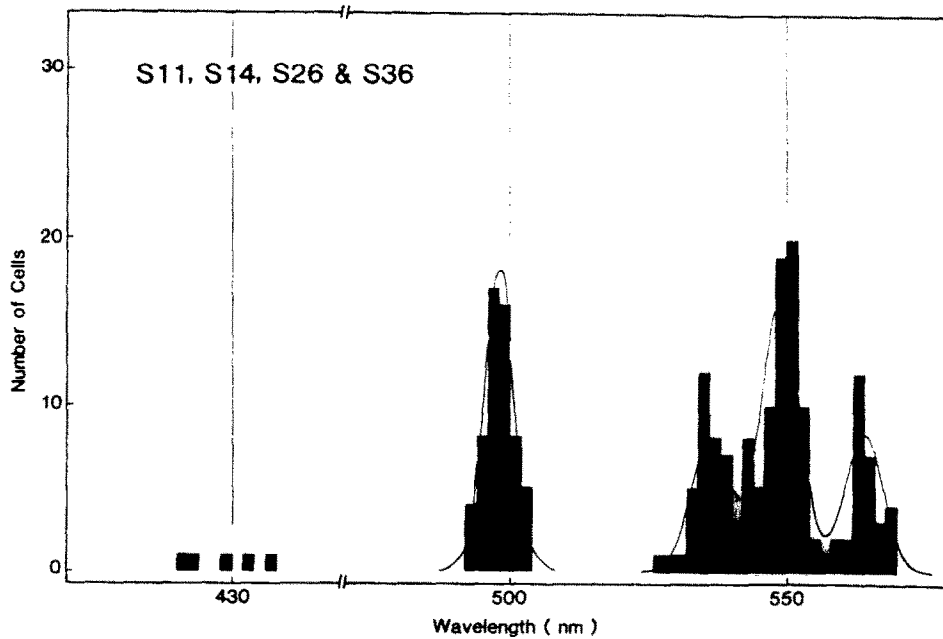


Fig. 3. Pooled distribution of λ_{\max} values from the four monkeys. The solid lines show (centre) the best-fitting normal distribution for the pooled rod records and (right) the distribution predicted by assuming three underlying normal distributions in the middle- to long-wave region; in the latter case the statistical model assumes that the P550 distribution is common to all four animals and that in a given monkey it is combined either with the P537 or the P563 distribution in the mixing proportion 0.5 (see text).

in the histograms of Figs 2 and 3. Our reasons for estimating λ_{\max} from the right-hand limb of the absorbance spectrum were that this limb includes the steepest part of the absorbance spectrum (so that absorbance varies rapidly with wavelength) and that this region of the absorbance spectrum is thought to be least subject to contamination by photoproducts and by wavelength-dependent scattering (MacNichol *et al.*, 1983).

All records were analysed with Dartnall's standard template for rhodopsin (Knowles and Dartnall, 1977), which had been placed with its λ_{\max} at 502 nm and then re-expressed in terms of the fourth root of wavelength. The reason for this procedure is that primate photopigments of varying λ_{\max} have almost the same shape when plotted against $\lambda^{1/4}$ (Barlow, 1982); and by using the same template for all cells we avoid the danger of creating an artificial bimodality, such as might be generated if two different templates, expressed in terms of wavenumber, were used for the two types of long-wave cone in the "deuteranomalous" retina (see Mollon *et al.*, 1984, for further discussion).

Except in the case of short-wave cones, records were excluded from further analysis if the peak value of the transverse absorbance was less than 0.01; or if the standard deviation of the 20 estimates of λ_{\max} was greater than 10 nm; or if there was a discrepancy of more than 10 nm between the estimate of λ_{\max} derived from the right-hand limb and an analogous estimate derived by fitting the template to both long- and short-wave regions of the absorbance spectrum. In

the case of short-wave cones, we included all cells that showed clear evidence of bleaching after a 5-min exposure to white light.

RESULTS

Behavioural tests

Results from the increment-threshold measurements are expressed as the \log_{10} ratio of the sensitivities measured at 540 and 640 nm. S11 and S26 showed relatively high sensitivity to the 640 nm test, exhibiting log ratios of 0.36 and 0.10 respectively, whereas S14 and S36 had lower long-wavelength sensitivity, exhibiting log ratios of 0.73 and 0.96.

The results from the colour vision tests are summarised in Fig. 1. In the Rayleigh match test all the subjects learned to discriminate various mixtures of red and green light from an equiluminant yellow. We therefore assume them to be trichromats or, more precisely, we assume that they have two different photopigments in the red-green spectral region (Jacobs, 1984). Each failed to discriminate some small range of mixture proportions from the yellow, and the midpoints of these ranges (i.e. the Rayleigh matches) are shown for each animal in the bottom panel of Fig. 1. Relative to the performance of normal human trichromats tested in the same situation, S14 and S36 required more red light at their match, whereas S11 and S26 required more green light at the match. The match locations for the members of each pair are very similar.

Table 1. Maximum-likelihood estimates of parameters of double-normal distributions fitted individually to four monkeys

| Monkey | <i>n</i> | $\hat{\lambda}_A, \hat{\lambda}_B$ (nm) (SE in brackets) | $\hat{\sigma}$ (nm) | \hat{p} | <i>R</i> Relative Goodness-of-fit |
|--------|----------|--|------------------------|-------------|---|
| S14 | 48 | 536.9 (0.7), 549.0 (1.1) | 3.45 (0.39) | 0.62 (0.08) | -309.5 |
| S36 | 22 | 535.7 (1.1), 548.8 (1.1) | 3.40 (0.52) | 0.47 (0.11) | -143.6 |
| S11 | 19 | 547.9 (1.4), 562.3 (2.6) | 4.47 (0.99) | 0.76 (0.12) | -128.3 |
| S26 | 54 | 549.6 (0.6), 564.5 (0.6) | 3.13 (0.32) | 0.52 (0.07) | -348.7 |
| | | | | | -930.1 |

From each of the two pairs distinguished by the preceding test, one animal was tested for wavelength discrimination. These results are shown in the standard format in the top panel of Fig. 1. The functions for both animals show the twin minima characteristically exhibited by human trichromats. The abilities of these two animals to discriminate wavelengths were closely similar over much of the spectrum. They differed, however, in that S14 was somewhat better than S11 at discrimination among the short wavelengths, whereas at long wavelengths (> 600 nm) S14 was very much poorer than S11.

On the basis of the behavioural results, we conclude that S11 and S26 resemble deuteranomalous human observers, whereas S14 and S36 resemble protanomalous human observers. Moreover, in their sensitivity ratios and colour discrimination, each of these pairs resembles one of the two types of squirrel monkey identified in the previous study.

Microspectrophotometric measurements

A total of 206 records satisfied the criteria specified in the Methods. Figure 2 shows for each animal the distribution of λ_{\max} values for individual receptors. In the middle- to long-wave region of the spectrum there is a very clear difference between the distributions for the two behaviourally "protanomalous" animals (S14, S36), on the one hand, and those for the behaviourally "deuteranomalous" animals (S11, S26), on the other. Relative to the protanomalous distributions, the deuteranomalous distributions are shifted to longer wavelengths. Some of the distributions are overtly bimodal, although none of the animals shows the clear separation of long- and middle-wave cones that would characterise a macaque (Bowmaker *et al.*, 1978; Bowmaker *et al.*, 1980; Hárosi, 1982; MacNichol *et al.*, 1983).

Analysis of middle- and long-wave cone distributions. A statistical analysis was undertaken in order to address directly the question raised in the Introduction. The strategy of the analysis had two stages. First, a rather complex model was found that provided an adequate description of the data in the middle- and long-wave region of the spectrum. Secondly, a simplifying hypothesis was introduced, and standard statistical techniques were used to check whether the more parsimonious model provided a fit that was as good (when random error had been taken into account) as that provided by the more complex hypothesis.

Except in the case of S11, where only a small

sample of records was obtained, none of the individual distributions could be adequately fitted by a single normal distribution. In view of this preliminary finding, and in view of the *prima facie* appearance of the distributions, the following model was tested. For each animal individually, a "double normal" distribution was fitted to the data. It was assumed that a proportion *p* of the observations arose from a single normal distribution with mean λ_A and standard deviation σ , and the remaining proportion $1 - p$ from a normal distribution with the same standard deviation but with mean λ_B , where $\lambda_B > \lambda_A$. The dashed lines in Fig. 2 show the individual double-normal distributions fitted numerically by maximum likelihood (Silvey, 1975; NAG, 1978). (The "likelihood" of a fitted model is expressed in terms of the plausibility of the observed data were the model true, and is defined as the product over *i* of ρ_i , where ρ_i denotes the height of the fitted curve at the *i*th observation. Maximising the likelihood can be shown to be equivalent to maximising a quantity *R*, which is equal to

$$2 \sum_i \log_e(\rho_i);$$

the formal use of *R* in comparing our two models is discussed below.) The parameters of the estimated distributions are given in Table 1 (where λ_A denotes the estimate of λ_A and so on).

From simple inspection of Fig. 2 it is apparent that this first model generally provides a good description of the data in the middle- to long-wave part of the spectrum; but for each animal a formal chi-square "absolute" goodness-of-fit test was carried out to see whether a double-normal distribution provided an adequate description of the data. This revealed that no data set departed significantly from the model.

It is clear from Table 1 that there is substantial homogeneity across monkeys. In particular, in view of our genetic theory (see Discussion) we investigated the simplifying hypothesis that the peaks of the component distributions align at three points and that the standard deviations are common across monkeys. The data also suggest testing the assumption that the "mixing proportion" is 0.5 for each pair of distributions. Estimates of the four resulting free parameters, denoted $\lambda_1, \lambda_2, \lambda_3$ and σ , were obtained by simultaneous maximum likelihood estimation across the four monkeys, parameters being constrained to be equal where relevant. The results for this second model are shown in Table 2, and the fitted curves are shown as continuous lines in Fig. 2.

The continuous curves of Fig. 2, although derived from a substantially simpler model, seem to provide almost as good a fit as those that are obtained when double-normal distributions are individually fitted to each animal. In formally assessing the adequacy of the simplifying hypothesis, it is inappropriate to compare directly the absolute goodness of fit, since substantial grouping of the data is required before calculating a χ^2 statistic and the absolute test is therefore rather insensitive to sparse outlying observations. A more sensitive test is provided by the measure R defined previously. Since R depends on the scale of measurement it measures only *relative* goodness-of-fit, but differences in R (the "likelihood ratio statistic") provide the most powerful test *between* competing models (Silvey, 1975). When a simpler model is fitted, the measure R always decreases, but, if the simple model is true, the decrease should be due to random variation and should be distributed approximately as a χ^2 variable with degrees of freedom equal to the extra parameters in the more complex model. For the present data the decrease in R is $(-930.1) - (-944.7) = 14.6$, while the simpler hypothesis has 12 fewer parameters. Thus $P \approx 0.25$, and we conclude that there is no evidence to reject the simple model in favour of individual double-normal fits.

When the middle- and long-wave data are pooled from all four monkeys, the histogram of Fig. 3 is obtained. The fitted curve in this figure provides a summary description of the four-parameter hypothesis suggested by the data.

From Fig. 2 it is apparent that a single low observation occurred near 535 nm in the case of each of the behaviourally deuteranomalous animals, S11 and S26; and it may be shown that there is only a 6% and a 0.6% chance, respectively, of observing such extreme observations if the simplified model were true. The high estimate of σ shown for S11 in Table 1 is due solely to the lowest observation, and in fact this observation is so influential as to make, for this monkey alone, the fit of the double-normal shape not a significant improvement on that of a single normal distribution. However, if the two outlying observations are removed from S11 and S26, the decrease in R is $(-887.0) - (-902.0) = 15.0$, showing that the conclusions are unaffected by these somewhat extreme data-points. The two outlying records in question satisfy our standard criteria (see Methods) and we therefore include them in the sample; our present judgement is that they most probably represent cases where a rod and a cone, lying in different planes,

chanced to be superimposed in such a way as not to be clearly distinguishable by the operator.

Derivation of mean absorbance spectra. The data suggest that the "protanomalous" and "deuteranomalous" retinae each contain two photopigments in the middle- to long-wave part of the spectrum, but the λ_{\max} values of the putative pigments are close together and the distributions of individual records are overlapping. How then can the mean absorbance spectrum best be estimated for a given pigment? Previously we have arbitrarily assigned the records to two groups according to the appearance of the overall distribution, and have then averaged the records within a group in order to derive a mean spectrum; this procedure will necessarily misassign a small number of cells and will thus produce a slight artificial separation of the mean spectra. The statistical analysis of the previous section permitted an improved analysis to be used for the present sample. For each animal we assumed that there were two underlying normal distributions of records with mean λ_{\max} values and standard deviations taken from Table 2. Each individual record was added to both averages with a weighting that depended on the ratio of the ordinates of the two normal distributions at the estimated λ_{\max} of that record. In most cases this meant that a cell contributed negligibly to one of the two averages, but cells near the middle of the overall distribution contributed to both averages. As previously, the raw records were not normalised before averaging; this procedure gives greater weight to records that have a higher absorbance and thus a better signal-to-noise ratio. In Fig. 4 we show for each animal these derived mean absorbance spectra for the two putative pigments. A pleasing feature is that in each case the relative absorbances at short wavelengths are very similar for the two pigments. This would be unlikely to be the case if any one of the derived pigments were an artifact of contaminated records, for the common sources of microspectrophotometric error reveal themselves by changes in short-wave absorbance (MacNichol *et al.*, 1983).

Short-wave cones. Short-wave receptors were rare, constituting less than 4% of all cones; this finding is in accord with previous microspectrophotometric results for *Saimiri* (Mollon *et al.*, 1984) and with microspectrophotometric and histological results for other primate species (Bowmaker *et al.*, 1978; Sperling, 1980; De Monasterio *et al.*, 1981; Dartnall *et al.*, 1983; Mansfield *et al.*, 1984). The filled squares in Fig. 5 show the mean absorbance spectrum for the five

Table 2. Maximim-likelihood estimates of parameters assuming double-normal fit to each monkey but with aligned means, common standard deviation and mixing proportions 0.5

| λ_1 (nm) | λ_2 (nm) | λ_3 (nm) | σ (nm) | R |
|---------------------|---------------------|---------------------|------------------|--------|
| 536.2 (0.7) | 548.8 (0.4) | 564.0 (0.6) | 3.51 (0.23) | -944.7 |

Note: values in brackets are standard errors.

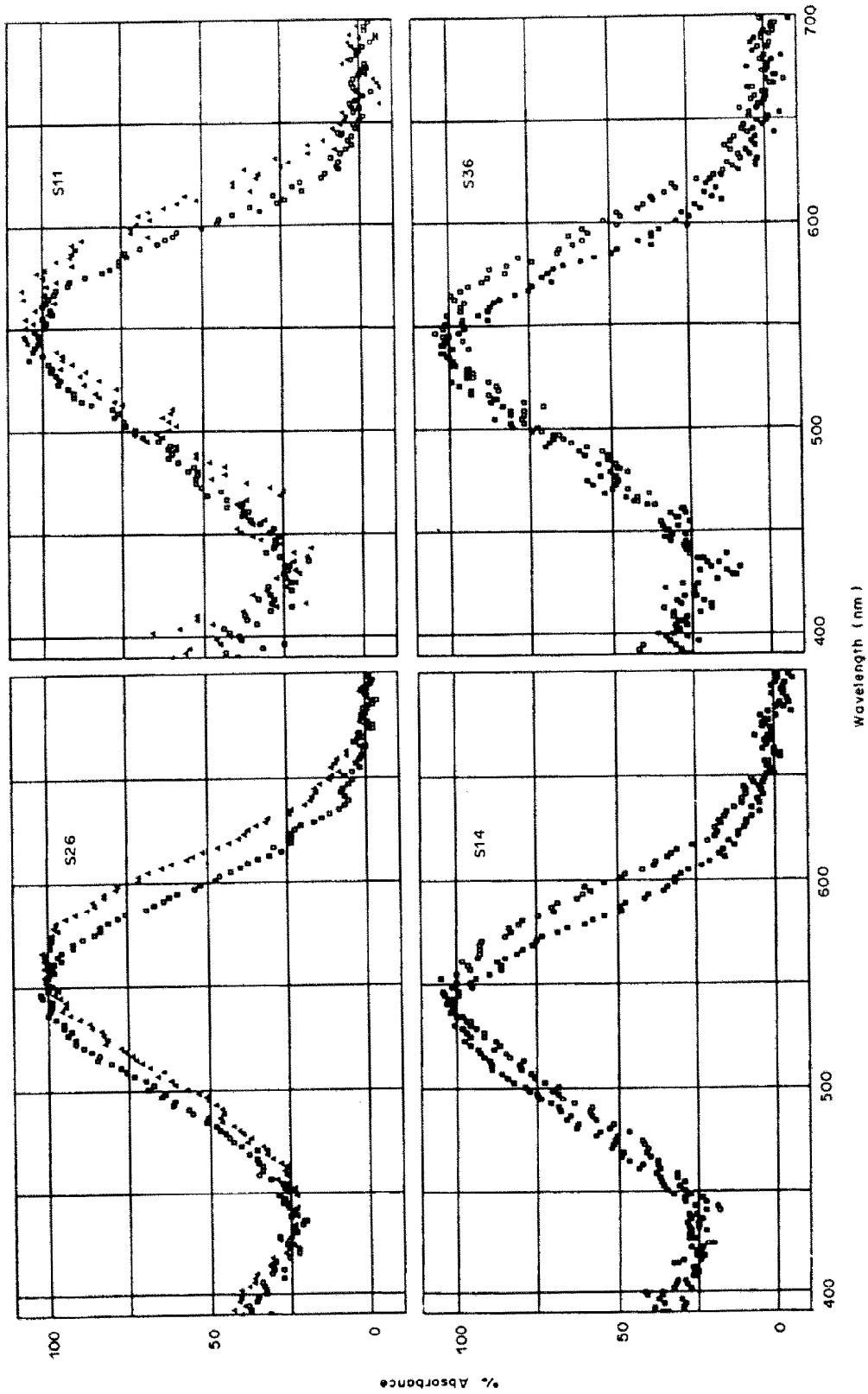


Fig. 4. Absorbance spectra for the two pigments thought to be present in the middle- to long-wave spectral region. Results are shown independently for individual animals, the behaviourally deuteranomalous animals above and the behaviourally protanomalous animals below. For details of the derivation of these spectra, see text.

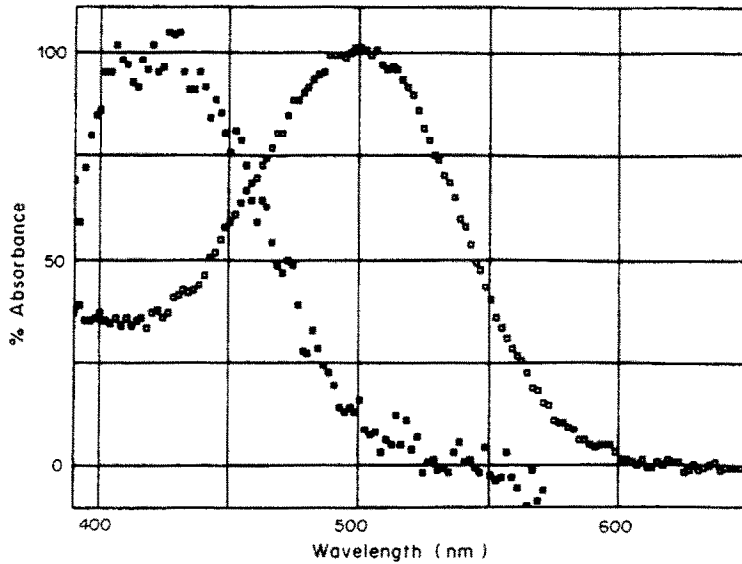


Fig. 5. Mean absorbance spectra for the short-wave cones (solid squares) and for the rods (open squares). Records have been pooled from the four animals to obtain these curves. Individual records were not normalised before averaging.

short-wave cones in this sample. The curve was derived by averaging the individual records and then normalising.

Rods. Records were analysed for a total of 58 rods. The solid lines superimposed on the rod histograms of Fig. 2 represent normal distributions fitted to the data for individual monkeys; the parameters of these distributions are listed in Table 3. Using an analysis analogous to that applied to the long- and middle-wave cones (see above), we have asked whether we can reject the simplifying hypothesis that a single rod distribution is common to all animals. If all the rod observations did come from the same normal distribution, then the decrease in fit between the more complex model (that of individual fits) and the simpler model should be due to random variation and should be distributed approximately as a χ^2 variable with degrees of freedom equal to the number of extra parameters in the more complex model. In fact the decrease in R is $-263.6 - (-275.2) = 11.6$, for which $P > 0.05$ for six degrees of freedom. Thus there is insufficient evidence to reject the hypothesis of homogeneity among monkeys, although admittedly the statistical significance does come close to the 5% level owing to the low λ_{\max} values observed for S14. However, a standard one-way analysis of variance, in

which variances are assumed equal among different monkeys, provides no evidence against the hypothesis of a homogeneous distribution. The parameters of the pooled rod distribution are listed in Table 3. A mean absorbance spectrum for the rods, derived by averaging records from all animals, is shown by the open squares in Fig. 5.

Mean absorbance at λ_{\max} . The absolute mean absorbance values at λ_{\max} , when equal weight is given to each animal, are 0.0145 for short-wave cones, 0.027 for rods, and 0.0215 for cones in the middle- to long-wave range part of the spectrum. These values are not definitive estimates of transverse densities, since some records (varying in number between animals) were secured from foveolar receptors lying nearly axial to the beam; but it is of interest that for any given animal the two types of cone in the red-green range gave closely similar mean absorbances. The values were 0.023, 0.023; 0.016, 0.016; 0.020, 0.018; and 0.029, 0.027 for monkeys S14, S36, S11 and S26 respectively.

DISCUSSION

Our independently obtained measurements—behavioural and microspectrophotometric—are in

Table 3. Maximum likelihood estimates of parameters of single normal distributions fitted (a) to rod data for each individual monkey and (b) to the pooled rod data from all monkeys

| Monkey | n | $\hat{\lambda}$ (nm) | $\hat{\sigma}$ (nm) | R |
|--------|-----|-------------------------|------------------------|--------|
| S11 | 18 | 497.8 (0.4) | 1.66 (0.28) | -69.2 |
| S14 | 6 | 495.8 (1.2) | 2.98 (0.86) | -30.1 |
| S26 | 18 | 498.5 (0.6) | 2.50 (0.42) | -84.1 |
| S36 | 16 | 498.1 (0.7) | 2.96 (0.52) | -80.2 |
| | | | | -263.6 |
| Pooled | 58 | 497.9 (0.3) | 2.60 (0.24) | -275.2 |

Note: values in brackets are standard errors.

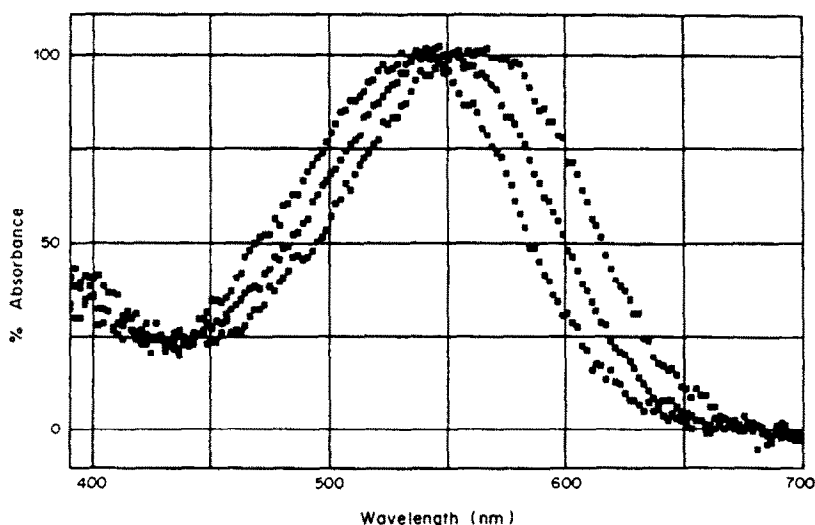


Fig. 6. The absorbance spectra of the three photopigments believed to occur in the middle- to long-wave spectral region. Either one or two of these pigments is thought to be available to any individual squirrel monkey. The three curves were obtained from those of Fig. 4. The central curve (P550) was obtained by averaging the corresponding curve from all four animals, equal weighting being given to each animal; the leftmost curve was obtained by similarly averaging the two middle-wave spectra of animals S14 and S36, and the rightmost curve was obtained by averaging the two long-wave spectra of S26 and S11.

good accord, and they confirm the existence of at least two types of trichromatic squirrel monkey. Our particular purpose was to ask whether monkeys of these two types share a common pigment in the red-green range. The statistical analysis of the microspectrophotometric results provides strong evidence for a shared pigment and supports the empirical generalisation suggested by our earlier study: *in the middle- to long-wave region of the spectrum three photopigments are potentially available to the squirrel monkey, and individual animals draw either one or two pigments from the set of three*. The three pigments have λ_{\max} values lying at approximately 536, 549 and 564 nm and Fig. 6 shows our best estimates of their normalised absorbance spectra. The curves of Fig. 6 have been derived from those of Fig. 4, each animal contributing to two of the curves. The relative absorbance at short wavelengths is closely similar for the three pigments; and the two pigments present in any individual animal exhibit the same absolute absorbance at λ_{\max} (see Results).

Hitherto we have used the terms "protanomalous" and "deuteranomalous" for the two types of monkey studied here. These terms acknowledge the similarities in behavioural test performance between the monkeys and the two commonest types of anomalous human observer. At the present time, the nature of anomalous trichromacy in man is less securely understood than is the colour vision of the squirrel monkey; but our work already shows that *Saimiri* cannot provide an exact model for human anomaly. We note in particular that (a) the Rayleigh matches of the "deuteranomalous" monkeys are not as extreme as is required by the conventional definition of deuteranomaly (Pokorny *et al.* 1979, p. 214) and (b) the

genetic basis of trichromacy is quite different in the two species.

We have suggested that in *Saimiri*, unlike man, there is only one genetic locus for a pigment in the middle- to long-wave part of the spectrum (Mollon *et al.*, 1984; Jacobs and Neitz, 1985). Three different alleles can occur at the locus. Each allele corresponds to a different opsin, which, when combined with 11-*cis* retinal, yields one of the absorbance spectra of Fig. 6. Further, we suppose that the single locus is on the X chromosome. Thus, a female monkey has the possibility of inheriting two different alleles from her two parents, whereas a male, having only one X chromosome, can inherit only one of the three alleles available in the population. An additional element in the theory is "X-chromosome inactivation" or "Lyonisation" (Lyon, 1962, 1972, 1974; Gartler and Riggs, 1983) which is known to occur during development of the female embryo: in any particular somatic cell, either the paternal or the maternal X-chromosome is inactivated; and the suppressed chromosome is thought to remain suppressed in all descendants of that cell. The importance of Lyonisation for the theory is that it prevents the occurrence of mixtures of photopigments within individual receptors: in the heterozygote only one of the two possible photopigments will be manufactured in any given one of those cells that are programmed to be long- or middle-wave cones. Finally, we assume that the nervous system of the heterozygote is plastic enough to exploit the presence of two classes of cone with different sensitivities in the red-green spectral region, so as to yield behavioural trichromacy.

In the two samples of squirrel monkeys that we have examined microspectrophotometrically (total

$n = 14$). we have identified five phenotypes—three types of dichromat and two types of trichromat. A sixth phenotype is clearly predicted by the particular genetic theory outlined above: there should exist trichromatic female monkeys that draw from the pool the alleles for the 536 nm and 564 nm pigments. Such animals would have a complement of photopigments very similar to those of Old World monkeys (Bowmaker *et al.*, 1978; Hárosi, 1982; MacNichol *et al.*, 1983) and quite similar to those of normal human trichromats (Dartnall *et al.*, 1983). This sixth phenotype remains to be identified by microspectrophotometry, but evidence for its existence is given by recent behavioural measurements (Jacobs, 1984).

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